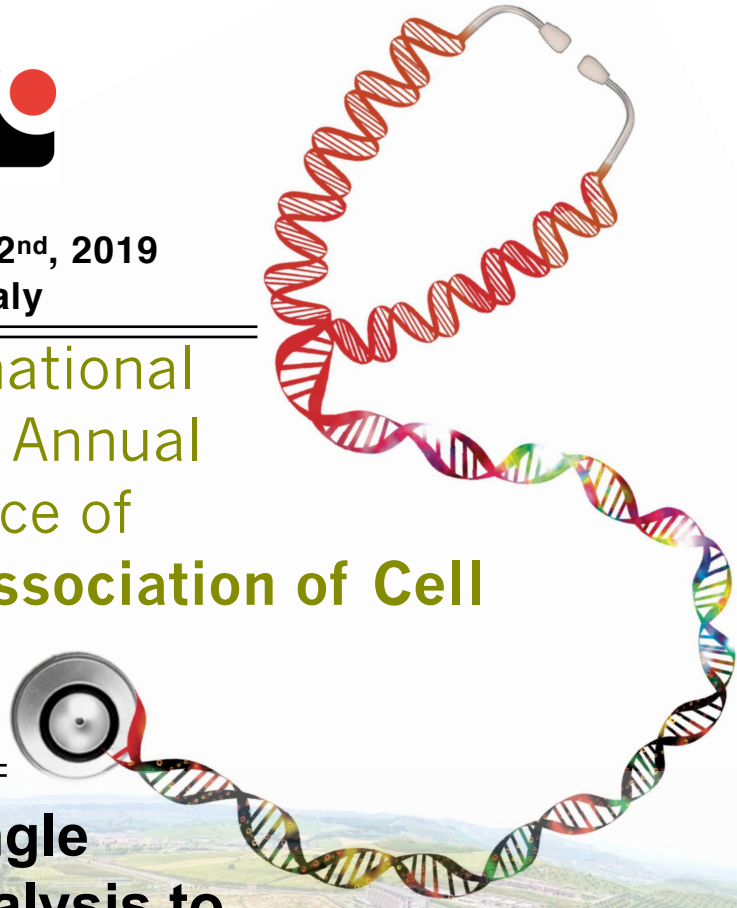
ASSOCIAZIONE ITALIANA DI COLTURE CELLULARI



October 1st - 2nd, 2019
Catanzaro, Italy

1st International and 32nd Annual Conference of **Italian Association of Cell Cultures** (AICC)

From Single Gene Analysis to Single Cell Profiling: A New Era for Genomic Medicine

Università Magna Græcia, Catanzaro
Aula Magna C
Level 1, building G

PROGRAMME AND ABSTRACT BOOK

UMG
Dubium sapientiae initium

1st International and 32nd Annual Conference

ASSOCIAZIONE ITALIANA DI COLTURE CELLULARI

P23. Oxidative stress induces *Wnt* canonical/non-canonical pathways modulation in colon cancer cell models

Catalano T.[1], Di Marcantonio M.C.[2], D'Amico E.[2], Moscatello C.[2], D'Agostino D.[2,4], Ferlazzo G.[3], Curia C.M.[2], Lanuti P.[4,5], Lattanzio R.[2,4], Aceto G.M.[2]

[1]Department of Clinical and Experimental Medicine, University of Messina, Italy; [2]Department of Medical, Oral and Biotechnological Sciences, 'G. d'Annunzio' University, Chieti, Italy; [3]Department of Human Pathology, University of Messina, Italy; [4]Center for Advanced Studies and Technology (CAST), Chieti, Italy; [5]Department of Medicine and Aging Sciences, University G. d'Annunzio of Chieti, Chieti, Italy
Email: tcatalano@unime.it

Background-aim. Increased reactive oxygen species (ROS) levels play critical roles in chronic inflammation, and predispose to colon carcinogenesis. Wnt signaling is essential for gut morphogenesis, tissue homeostasis and self-renewal, but its aberrant activation may drive the colorectal cancer (CRC). The ROS production seems to induce the Wnt/β-Catenin pathways, but the molecular mechanisms involved in CRC progression are still undefined. To evaluate the molecular relationship among oxidative stress and canonical/non-canonical *Wnt* pathways, we analyzed the response to ROS exposure in CRC cell lines with different *Wnt* signaling behaviour.
Methods. HCT116 (MSI) and SW480 (MSS) cells were treated with H_2O_2 [2 mM and 10 mM] for 15'and 30'. We assayed cell viability by MTS and cell cycle by FACS. Gene expression was evaluated by SYBR Green qRT-PCR, and protein expression was analyzed by IHC. Statistical analysis was performed by T-test (p value<0.05).
Results. MTS revealed different inhibition rates of cell growth at H_2O_2 concentrations. Acute stress induced by H_2O_2 [2mM] up-regulated gene expression of canonical LRP6 and LEF1, and non canonical ROR2 and JUN/AP1 molecules in SW480, while reduced ROR2 and LRP6 expression in HCT116. Both pathways showed a dose dependent increase in SW480, at H_2O_2 [10mM]. In HCT116 down-regulated gene expression of *APC*, *LRP6*, *LEF1*, and *p65-NFkB* was dependent on treatment time, in opposition to non-canonical *ROR2*. *MUTYH*, *OGG1*, *NRF2*, *COX2* and *JUN/AP1* expression significantly increased. H_2O_2 treatment induced FZD6 protein expression in HCT116 cytoplasm and E-cadherin protein expression in SW480 cytoplasm, while beta-catenin increased in both cell lines. Intriguingly we relieved a *de novo* APC expression in both cell lines cytoplasm. FACS analysis of cell cycle showed time dependent changes: upon H_2O_2 [2mM] treatment at 15', SW480 increased in G1 and G2 and decreased in S, whereas HCT116 increased in G1 and slightly reduced in G2; after 30', SW480 enhanced in G1 and S, and reduced in G2 while HCT116 diminished in G1 and increased in S/G2.
Conclusions. In MSI and MSS CRC cells, oxidative stress differently affects the *WNT* pathways at gene and protein expression levels. Our results could unravel a new scenario for innovative CRC therapeutic approaches.